A Research Note

THE ROLE OF LEAN AND ADIPOSE TISSUE ON THE FORMATION OF NITROSOPYRROLIDINE IN FRIED BACON

INTRODUCTION

FRIED BACON has been reported to contain nitrosopyrrolidine (NO-Pyr) in concentrations as high as 108 ppb (Crosby et al., 1972; Fazio et al., 1973; Pensabene et al., 1974; Sen et al., 1973). This is of concern from a public health standpoint since NO-Pyr has been shown to be carcinogenic when administered to rats (Druckrey et al., 1967; Greenblatt and Lijinsky, 1972a; Greenblatt et al., 1973) and mice (Greenblatt and Lijinsky, 1972b). Some investigators have reported a larger concentration of NO-Pyr in the cooked-out fat than in the fried bacon (Fazio et al., 1973; Pensabene et al., 1974; Sen et al., 1973). This may be accounted for by the fact that NO-Pyr is very fat-soluble and partitions into the fat phase, but the possibility should be considered that larger concentrations of nitrosamine are formed in the adipose tissue. The role of lean and adipose tissues of bacon as precursors for NO-Pyr was investigated and the results reported herein.

EXPERIMENTAL

BACON was purchased from three different manufacturers a few days after processing. A portion of the bacon was visually separated into adipose and lean components. Three brands of Canadian bacon (back bacon); ham, including a cured pork shoulder; and breakfast beef (bacon-like products) were purchased at local retail markets. The samples were fried in a preheated Presto teflon-coated electric frying pan (Model PC4AT) for 6 min at a calibrated thermostat setting of 177°C. The lean portions of bacon, ham and Canadian bacon samples were fried in Crisco, a hydrogenated vegetable oil product, under the same conditions.

The samples were assayed for dimethylnitrosamine, methylethylnitrosamine, diethylnitrosamine, nitrosopiperidine, nitrosopyrrolidine and nitrosomorpholine. The fried bacon samples were analyzed using the multidetection method as described by Fazio et al. (1971) and the drippings as reported by White et al. (1974) except for a modification of the column chromatographic clean-up step in which a 4-g layer of Silica Gel was used on top of 1g of Florisil acidified with 1 ml of 6N HCl. The sample was washed with 200 ml of CH_2Cl_2/pentane (1/1) and eluted with 125 ml of ether/CH_2Cl_2 (1/5). The nitrosamines were determined by GLC using an alkali flame ionization detector (Howard et al., 1970). Concentrations as low as 0.5 ppb nitrosamine could be detected. Only NO-Pyr was found in confirmable concentrations. The average recovery of 20 ppb added NO-Pyr was 75% in the fried samples and 50% in the drippings. When a peak was observed at the same retention time as an authentic sample of NO-Pyr, its identity was confirmed by mass spectrometry in the peak-matching mode using the parent peak of m/e 100.06366 at a resolution of 1 in 12,000. The GLC and GLC-MS systems and conditions have been described previously (Pensabene et al., 1974).

RESULTS & DISCUSSION

THE RESULTS of analyses for NO-Pyr in fried whole bacon and in the lean and adipose tissue portions fried separately are shown in Table 1. Results from samples of three different producers were similar. In the case of whole bacon, NO-Pyr was found in the fried product and its drippings. No NO-Pyr was found in uncooked adipose tissue which had been separated from the lean portion of raw bacon. However, after frying, more NO-Pyr was found in the cooked out fat than in the solid residue remaining after frying. The lean portion did not contain NO-Pyr when uncooked, pan-fried alone, or when fried in Crisco under the same conditions as the bacon adipose tissue. No NO-Pyr was detected in heated Crisco alone or in Crisco remaining after the lean was fried in it. It appears that NO-Pyr is derived from the adipose tissue and not the lean portion of bacon. Some components in the adipose tissue therefore must serve as precursor(s) for NO-Pyr. While the exact mechanism for NO-Pyr formation in bacon is not known, several pathways have been proposed, as shown in Figure 1.

Table 1—Effect of lean and adipose tissue on nitrosopyrrolidine formation in fried bacon

	Nitrosopyrrolidine,[a] ppb		
	Samples		
Product	A	B	C
Whole bacon	2	28	13
Drippings	6	24	22
Fat residue	5	n.d.	11
Drippings	14	58	24
Lean (in Crisco)	n.d.[b]	n.d.	n.d.
Drippings (Crisco)	n.d.	n.d.	n.d.
Fat—unfried	n.d.	n.d.	n.d.
Lean—unfried	n.d.	n.d.	n.d.

[a] Confirmed by high resolution mass spectrometry
[b] n.d. = none detected

Bills et al. (1973) have reported NO-Pyr can be formed in the highest yield from nitrosoproline and in lesser quantities from nitrite reacted with pyrrolidine, spermidine, proline and putrescine at 170°C. In addition, NO-Pyr has been formed by decarboxylating nitrosoproline at different temperatures with the maximum formation occurring at 185°C, close to the recommended temperature for frying bacon, 177°C (Fiddler et al., 1973; Pensabene et al., 1974). Therefore, proline indirectly appears to be the most probable of the above-mentioned precursors. The amount of free proline in bacon and ham is approximately the same (Lakritz, 1973). The fact that NO-Pyr is found only in cooked bacon and not in other cured pork products suggests that NO-Pyr is not formed via a simple mechanism. Fazio et al. (1973) found no NO-Pyr in fried ham or Canadian bacon and theorized that the nitrosamine, being fat soluble, is protected from volatilization during frying and is retained on the fried bacon strips. We examined ham, Canadian bacon (back bacon), and beef bacon-like products. Samples of ham and Canadian bacon were fried in Crisco. Nitrosopyrrolidine was not found in either the fried product, its cooked-out fat, or the Crisco in which it was fried. However, it is possible that the composition of pork belly is different than either the ham, shoulder or loin portions, particularly with respect to the amount of collagen or connective tissue present. Collagen is known to contain large concentrations of bound proline and hydroxyproline and has recently been claimed to produce NO-Pyr at elevated temperatures in a model system (Huxel et al., 1973). Nitrosopyrrolidine could form from the action of nitrite on collagen itself or its pyrolytic decomposi-

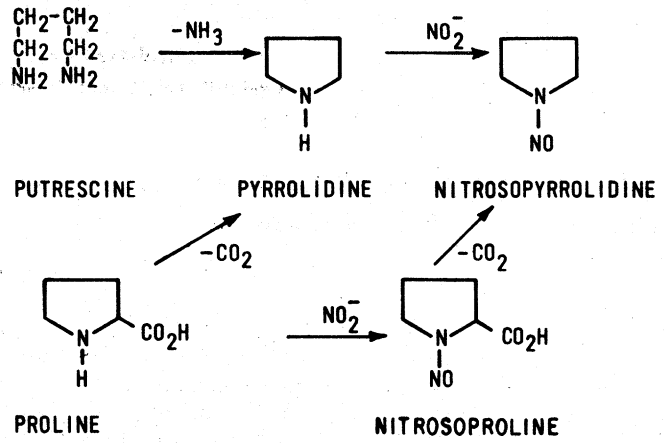

Fig. 1—Possible pathways for the formation of nitrosopyrrolidine in fried bacon.

tion products. Collagen might also hydrolyze to provide a higher concentration of free proline than is normally available for nitrosation in nonbelly cuts of pork. While the latter explanation is the most reasonable, the exact role of collagen and other components present in bacon adipose tissue regarding nitrosamine formation needs to be investigated.

(Precautions should be exercised in the handling of nitrosamines, since they are potential carcinogens.)

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Reference to brand or firm names does not constitute endorsement by the U.S. Dept. of Agriculture over others of a similar nature not mentioned.